ELSEVIER

www.elsevier.com/locate/farmac

Il Farmaco 56 (2001) 665–675

IL FARMACO

Synthesis and antibacterial activity of 2-substituted 6-fluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids

Jae-Chul Jung, Shin Baek, Oee-Sook Park *

Department of Chemistry, College of Natural Sciences, Chungbuk National, University, Cheongju 361-763, Chungbuk, South Korea

Received 19 October 2000; accepted 28 March 2001

Abstract

A series of 2-substituted 6-fluoro-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids was prepared and evaluated for antibacterial activity. The 6-fluoro-2-methyl-1-prenyl-1,4-dihydro-7-(3,5-dimethylpiperazinyl)-4-oxo-3-quinolinecarboxylic acid (**14f**) exhibited the most potent antibacterial activity against Gram-positive bacteria among the total 32 derivatives. The synthetic strategies involve the use of well known keto ester condensation of benzoyl chloride and reductive cyclization of intermediates (**4a**–**d**) to afford 4-hydroxy-1,2-dihydro-2-oxo-quinoline derivatives (**5a**,**b**) or 1-hydroxy-1,4-dihydro-4-oxo-quinoline derivatives (**6a**,**b**).

Keywords: 2-Substituted 6-fluoro quinolones; Antibacterial activity

1. Introduction

Quinolone antibacterial agents have become a significant class of therapeutically useful compounds. Since nalidixic acid, which shows a moderate activity on Gram-negative bacteria, was discovered by Lesher in 1962 [1], a large number of its analogues have been synthesized and evaluated. These compounds have been shown to inhibit bacterial growth by binding to the A subunit of DNA gyrase as well as the mammalian topoisomerase II [2,3]. Structure–activity relationships (SAR) developed through systematic modification of various ring positions have identified certain optimum substitution patterns. Especially active were analogues with a cyclopropyl group at the N-1, fluorine at the C-6 and the C-8 and a cyclic amine at the C-7 [4,5]. Although other quinolone ring positions have also been modified with varying degrees of success, very few modifications have been carried out at the C-2 [6].

Most of the quinolones currently on the market or under development have only moderate activity against many Gram-positive cocci including *Staphylococci* and *Streptococci*. This insufficient activity has not only limited their use in infections caused by these organisms, such as respiratory tract infections [7], but has also been believed to be one of the reasons for the rapidly developing quinolone resistance [8]. Therefore, recent efforts have been directed toward the synthesis of new quinolones that can provide improved Gram-positive antibacterial activity, while retaining the good Gram-negative activity of ciprofloxacin [9]. Gram-positive and Gram-negative bacteria differ remarkably in the features of their cell walls. The cell wall of Gram-positive bacteria is more lipophilic than that of Gram-negative bacteria.

Considering the above facts, we have focused our attention on the introduction of the substituent at the C-2 and prenyl group at the N-1 of the quinolone in order to find potent broad-spectrum antibacterial agents which display strong Gram-positive activity. We believed that the prenyl group would be an excellent choice for our purpose. First of all, this group can be readily introduced to the corresponding quinolone in a simple manner. Secondly, it enhances the lipophilicity of the quinolone.

In this paper, we describe the design and a versatile synthetic method for preparing 2-substituted quinolones and 4-hydroxyquinolin-2(1*H*)-one compounds. We also report herein the antibacterial activity data of the new quinolones.

* Corresponding author.
E-mail address: ospark@cbucc.chungbuk.ac.kr (O.-S. Park).

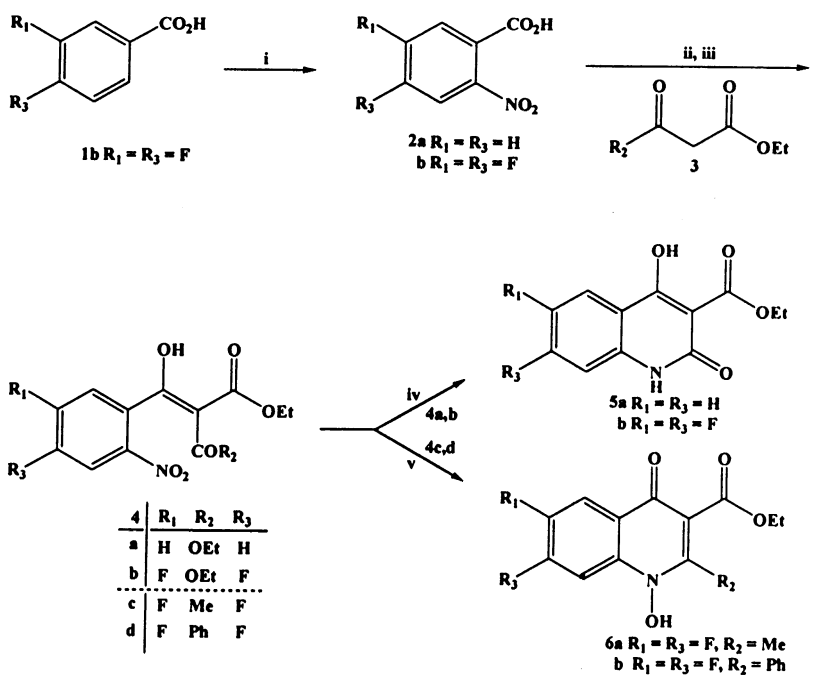
0014-827X/01/$ - see front matter
PII: S0014-827X(01)01112-0

2. Chemistry

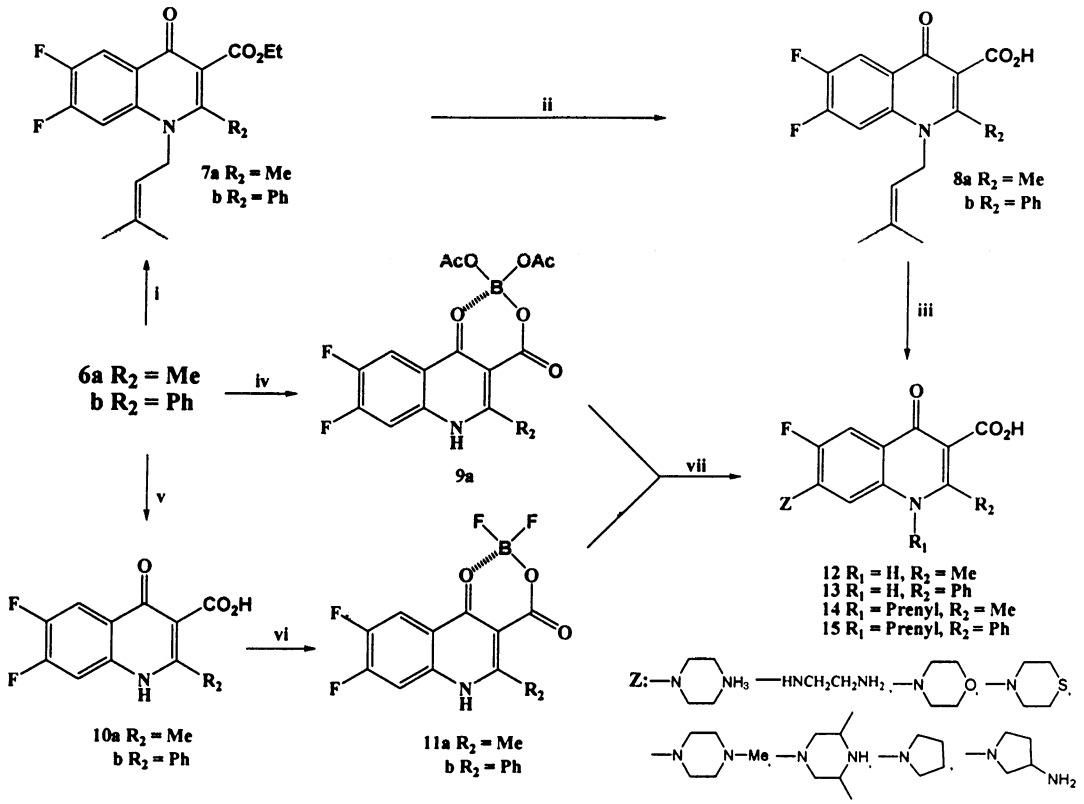
The synthesis of 1-hydoxy-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid derivatives (**6a**,**b**) and 4-hydroxy-1,2-dihydro-2-oxo-3-quinolinecarboxylic acid derivatives (**5a**,**b**) is outlined in Scheme 1. We have used 2-nitro-benzoic acids and 3,4-difluorobenzoic acid as starting materials. 3,4-Difluorobenzoic acid was nitrated to give 4,5-difluoro-2-nitrobenzoic acids (**2b**) in 86% yields. Acids **2a**,**b** were reacted with thionyl chloride to give acid chlorides, which were subsequently treated with the anion of ethyl acetoacetate, phenyl acetoacetate or diethyl malonate to give keto esters **4a**–**d** in excellent yields. In this condensation reaction, we found that the magnesium ethoxide was the most effective among bases such as sodium ethoxide, sodium hydride and potassium *tert*-butoxide. The keto diesters **4a**,**b** were smoothly transformed into compounds **5a**,**b** by reductive ring cyclization with sodium borohydride under alkaline condition, Whereas, the diketo esters **4c**,**d** were converted into compounds **6a**,**b** by a mild catalytic hydrogenation over palladium-on-charcoal in ethanol at room temperature.

The prenylation of 6,7-difluoro-1-hydroxy-2-(methyl or phenyl)-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid ethyl esters (**6a**,**b**) with prenyl bromide and anhydrous potassium carbonate in *N*,*N*-dimethylformamide at 80–90°C directly afforded 6,7-difluoro-2-(methyl or phenyl)-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecaboxylic acid ethyl esters (**7a**,**b**), which were hydrolyzed with aqueous sodium hydroxide or lithium hydroxide and then acidified with 3*N* hydrochloric acid to give the 6,7-difluoro-2-(methyl or phenyl)-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecaboxylic acids (**8a**,**b**) in good yields. The desired substituted 6-fluoro-2-(methyl or phenyl)-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids **14a**–**h** and **15a**–**h** were synthesized by the replacement of the C-7 fluorine atom with the appropriate amine nucleophiles according to the reported method [10,11].

On the other hand, the compounds **6a**,**b** were hydrolyzed with aqueous sodium hydroxide or to give the 6,7-difluoro-2-(methyl or phenyl)-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids (**10a**,**b**). In order to activate the C-7 position, the compound **6a** was treated with boric acid in acetic anhydride to produce diacetoxy[[[6,7-difluoro-2-methyl-1,4-dihydro-4-oxo-quinolin-3-yl]carbonyl]oxy]-borane (**9a**) in moderate yield [12]. Acids **10a,b** were also treated with tetrafluoroboric acid in water to yield difluoro[[[6,7-difluoro-2-(methyl or phenyl)-1,4-dihydro-4-oxo-quinolin-3-yl]carbonyl]-oxy]boranes (**11a**,**b**) in moderate yields [13]. The borate complex **9a** and **11a**,**b** were also reacted with a variety of amines in acetonitrile at room temperature and then hydrolyzed with 2*N* hydrochloric acid to yield-7-substituted 6-fluoro-2-(methyl or phenyl)-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids **12a**–**h** and **13a**–**h** (Scheme 2). A summary of the physical properties and analytical data for the 2-substituted quinolone compounds (**12a**–**h, 13a**–**h, 14a**–**h** and **15a**–**h**) are presented in Table 1.



Scheme 1. Reagents and reaction conditions: (i) HNO_3/H_2SO_4, 10°C, 1 h; (ii) $SOCl_2$, urea/toluene, 100°C, 3 h; (iii) Mg, EtOH, CCl_4/toluene, room temperature, 30 min; (iv) $NaBH_4$, Pd–C, NaOH(aq), 1,4-dioxane, room temperature, 30 min; (v) H_2, Pd–C, EtOH, room temperature, 3 h.



Scheme 2. Reagents and reaction conditions: (i) prenyl bromide, K_2CO_3/DMF, 80–90°C, 6 h; (ii) 2N NaOH reflux 3 h, or LiOH/THF, room temperature, 18 h; (iii) amines/pyridine, 100°C; (iv) H_3BO_3/Ac_2O, 100°C, 3 h; (v) 2N NaOH, reflux, 3 h, 10% HCl; (vi) HBF_4/H_2O, 100°C, 3 h; (vii) amines/acetonitrile, room temperature.

3. Biological activity

Compounds **12a–h, 13a–h, 14a–h** and **15a–h** were evaluated for in vitro antibacterial activity against standard organisms [Gram-positive bacteria: *Streptococcus pyogenes* 308 A, *Streptococcus pyogenes* 77 A (Sp), *Streptococcus faccium* (Sf), *Staphylococcus aureus* (Sa); Gram-negative bacteria: *Escherichia coli* (Ec), *Pseudomonas aeruginosa* 9027, *Pseudomonas aeruginosa* 1771 M (Pa), *Salmonella typhimurium* (St), *Klebsiella oxytoca* 1082 E (Ko), *Klebsiella aerogenes* 1522 E (Ka), *Enterobacter cloacae* P 99 (Ec P 99)]. Among the 2-substituted quinolone derivatives, the 6-fluoro-1-prenyl-2-methyl-1,4-dihydro-7-(3,5-dimethylpiperazinyl)-4-oxo-3-quinolinecarboxylic acid (**14f**) and fifteen compounds (**14a–e**, **14g–h** and **15a–h**) of 2-substituted quinolone derivatives showed better antibacterial activities against Gram-positive bacteria than Gram-negative bacteria. The compound **14f** exhibited the most potent antibacterial activity against Gram-positive bacteria among the total thirty-two derivatives.

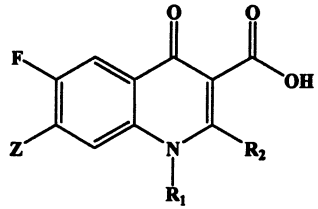
We propose that *N*-prenyl 2-substituted (methyl or phenyl) quinolone derivatives improve antibacterial activity for the Gram-positive bacteria in vitro, presumably owing to the increased lipophilicity and enhanced self-association [14].

The minimum inhibitory concentrations (MICs) of these compounds and norfloxacin against several Gram-positive bacteria and Gram-negative bacteria in vitro are listed in Table 2.

4. Experimental

Reactions requiring anhydrous conditions were performed with the usual precautions for rigorous exclusion of air and moisture. Tetrahydrofuran and diethyl ether were distilled from sodium benzophenone ketyl prior to use. Thin layer chromatography (TLC) was performed on precoated silica gel 60 F_{254} plates from EM reagents and visualized with 254-nm UV light or ceric sulfate-ammoniummolybdate-sulfuric acid spray. Flash chromatography was carried out on silica gel 60 (E.M. Merck, particle size 0.040–0.063 mm, 230–400 mesh ASTM). ^1H NMR and ^{13}C NMR spectra were recorded on a Brucker DPX 300 at 300 and 75.47 MHz, respectively. The chemical shifts were reported in parts per million (ppm) downfield from tetramethylsi-

Table 1
Physical data for 2-substituted quinolone derivatives



Comp.	R_1	R_2	*Z*	Method [a]	Yield [b] (%)	M.p. (°C)	^{1}H NMR (DMSO-d_6)
12a	H	Me	piperazine	B	68	216–217	13.62 (br s, 1H), 10.64 (br s, 1H), 9.27 (br s, 1H), 8.25 (d, 1H, $J = 7.76$ Hz,), 8.03 (d, 1H, $J = 7.31$ Hz), 3.64–3.33 (m, 4H), 2.91–3.24 (m, 4H), 2.38 (s, 3H)
12b	H	Me	ethylenediamine	C	65	237–238	12.56 (br s, 1H), 9.08 (br s, 1H), 7.81 (d, 1H, $J = 7.76$ Hz), 6.98 (d, 1H, $J = 7.02$ Hz), 6.22–5.97 (m, 3H), 3.03–2.86 (m, 4H), 2.58 (s, 3H)
12c	H	Me	1-methylpiperazine	B	67	246	13.81 (br s, 1H), 10.92 (br s, 1H), 8.28 (d, 1H, $J = 7.88$ Hz), 8.17 (d, 1H, $J = 7.71$ Hz), 3.81–3.55 (m, 4H), 3.30–2.86 (m, 4H), 2.44 (s, 3H), 2.29 (s, 3H)
12d	H	Me	morpholine	B	48	240 (dec) [c]	13.62 (br s, 1H), 11.91 (br s, 1H), 7.73 (d, 1H, $J = 8.02$ Hz), 6.67 (d, 1H, $J = 6.92$ Hz), 3.71–3.56 (m, 4H), 3.22–2.90 (m, 4H), 2.54 (s, 3H)
12e	H	Me	thiomorpholine	A	55	226–227	13.21 (br s, 1H), 12.56 (br s, 1H), 7.68 (d, 1H, $J = 7.96$ Hz), 6.59 (d, 1H, $J = 7.07$ Hz), 3.64–3.41 (m, 4H), 3.30–2.93 (m, 4H), 2.46 (s, 3H)
12f	H	Me	3,5-dimethylpiperazine	B	66	233–234	12.84 (br s, 1H), 11.06 (br s, 1H), 9.86 (br s, 1H), 7.42 (d, 1H, $J = 7.82$ Hz), 6.61 (d, 1H, $J = 7.02$ Hz), 3.24–2.80 (m, 6H), 2.35 (s, 3H), 1.03 (s, 3H), 0.95 (s, 3H)
12g	H	Me	pyrrolidine	C	62	218	11.98 (br s, 1H), 10.91 (br s, 1H), 7.61 (d, 1H, $J = 7.81$ Hz), 6.48 (d, 1H, $J = 6.54$ Hz), 3.26–2.90 (m, 4H), 2.38 (s, 3H), 1.88–1.59 (m, 4H)
12h	H	Me	3-aminopyrrolidine	C	56	196–197	12.41 (br s, 1H), 10.26 (br s, 2H), 9.21 (br s, 1H), 7.46 (d, 1H, $J = 8.34$ Hz), 6.34 (d, 1H, $J = 7.09$ Hz), 3.91–3.72 (m, 1H), 3.12–3.01 (m, 4H), 2.31 (s, 3H), 2.11–1.82 (m, 2H)
13a	H	Ph	piperazine	B	70	204	13.66 (br s, 1H), 10.13 (br s, 1H), 9.56 (br s, 1H), 8.12 (d, 1H, $J = 8.05$ Hz), 8.09 (dd, 1H, $J = 7.90$ Hz, $J = 7.92$ Hz), 7.73–7.48 (m, 5H), 3.68–3.45 (m, 4H), 3.21–2.98 (m, 4H)
13b	H	Ph	ethylenediamine	C	61	205–206	12.11 (br s, 1H), 9.42 (br s, 2H), 8.93 (br s, 2H), 7.78–7.44 (m, 6H), 6.32 (d, 1H, $J = 7.30$ Hz), 3.15–2.94 (m, 4H)
13c	H	Ph	1-methylpiperazine	A	66	209–210	13.22 (br s, 1H), 10.13 (br s, 1H), 7.78–7.47 (m, 6H), 6.64 (d, 1H, $J = 7.70$ Hz), 3.32–3.05 (m, 4H), 2.48–2.31 (m, 4H), 2.29 (s, 3H)
13d	H	Ph	morpholine	A	52	184–185	12.93 (br s, 1H), 9.45 (br s, 1H), 7.66–7.51 (m, 6H), 6.59 (d, 1H, $J = 7.06$ Hz), 3.76–3.59 (m, 4H), 3.32–2.98 (m, 4H)
13e	H	Ph	thiomorpholine	A	48	203–204	13.11 (br s, 1H), 9.12 (br s, 1H), 7.67–7.36 (m, 6H), 6.62 (d, 1H, $J = 7.07$ Hz) 3.64–3.41 (m, 4H), 3.30–2.93 (m, 4H)
13f	H	Ph	3,5-dimethylpiperazine	A	60	223–244	12.43 (br s, 1H), 10.53 (br s, 2H), 7.67–7.25 (m, 6H), 6.58 (d, 1H, $J = 7.47$ Hz), 3.34–2.89 (m, 6H), 1.10 (s, 3H), 1.05 (s, 3H)
13g	H	Ph	pyrrolidine	B	69	174	12.78 (br s, 1H), 11.66 (br s, 1H), 7.78–7.33 (m, 6H), 6.56 (d, 1H, $J = 6.76$ Hz), 3.20–2.79 (m, 4H), 1.91–1.78 (m, 4H)
13h	H	Ph	3-aminopyrrolidine	B	64	229–230	12.76 (br s, 1H), 10.14 (br s, 1H), 9.21 (br s, 2H), 7.76–7.29 (m, 6H), 6.72 (d, 1H, $J = 7.79$ Hz), 3.89–3.62 (m, 1H), 3.20–3.07 (m, 4H), 2.16–1.85 (m, 2H)
14a	prenyl	Me	piperazine	A	60	194–195	12.82 (br s, 1H), 10.81 (br s, 1H), 8.07 (d, 1H, $J = 7.66$ Hz), 7.86 (d, 1H, $J = 7.42$ Hz), 5.62 (t, 1H, $J = 7.75$ Hz), 4.76–4.54 (m, 2H), 3.67–3.42 (m, 4H), 3.18–2.76 (m, 4H), 2.21 (s, 3H), 1.62 (s, 3H), 1.52 (s, 3H)

Table 1 (*Continued*)

Comp.	R_1	R_2	*Z*	Method [a]	Yield [b] (%)	M.p. (°C)	^{1}H NMR (DMSO-d_6)
14b	prenyl	Me	ethylenediamine	B	59	228 (dec) [c]	12.48 (br s, 1H), 10.23 (br s, 1H), 9.42 (br s, 2H), 7.74 (d, 1H, $J = 7.81$ Hz), 6.88 (d, 1H, $J = 7.16$ Hz), 6.25 (m, 3 H), 5.67 (t, 1H, $J = 7.71$ Hz), 4.86–4.71 (m, 2H), 3.08–2.91 (m, 4H), 2.50 (s, 3H), 1.67 (s, 3H), 1.54 (s, 3H)
14c	prenyl	Me	1-methylpiperazine	A	68	212	13.77 (br s, 1H), 8.09 (d, 1H, $J = 7.79$ Hz), 8.23 (d, 1H, $J = 7.67$ Hz), 5.59 (t, 1H, $J = 7.61$ Hz), 4.81–4.61 (m, 2H), 3.61–3.48 (m, 4H), 3.27–2.72 (m, 4H), 2.48 (s, 3H), 2.33 (s, 3H), 1.58 (s, 3H), 1.52 (s, 3H)
14d	prenyl	Me	morpholine	A	62	242–243	13.28 (br s, 1H), 7.85 (d, 1H, $J = 7.91$ Hz), 6.72 (d, 1H, $J = 6.84$ Hz), 7.71 (t, 1H, $J = 7.84$ Hz), 4.77–4.60 (m, 2H), 3.62–3.47 (m, 4H), 3.12–2.81 (m, 4H), 2.34 (s, 3H), 1.69 (s, 3H), 1.58(s, 3H)
14e	prenyl	Me	thiomorpholine	B	61	238–239	13.16 (br s, 1H), 7.67 (d, 1H, $J = 7.84$ Hz), 6.85 (d, 1H, $J = 7.07$ Hz), 5.63 (t, 1H, $J = 7.61$ Hz), 4.82–4.69 (m, 2H), 3.61–3.42 (m, 4H), 3.18–2.76 (m, 4H), 2.43 (s, 3H), 1.71 (s, 3H), 1.57 (s, 3H)
14f	prenyl	Me	3,5-dimethylpiperazine	B	71	220–221	12.77 (br s, 1H), 10.95 (br s, 1H), 7.51 (d, 1H, $J = 7.88$ Hz), 6.72 (d, 1H, $J = 7.11$ Hz), 5.72 (t, 1H, $J = 7.70$ Hz), 4.65–4.32 (m, 2H), 3.31–2.99 (m, 6H), 2.41 (s, 3H), 1.58 (s, 3H), 1.49 (s, 3H), 1.08 (s, 3 H), 0.91 (s, 3H)
14g	prenyl	Me	pyrrolidine	C	65	229–231	11.98 (br s, 1H), 7.61 (d, 1H, $J = 7.94$ Hz), 6.48 (d, 1H, $J = 6.89$ Hz), 5.73 (t, 1H, $J = 7.74$ Hz), 4.78–4.50 (m, 2H), 3.26–2.90 (m, 4H), 2.38 (s, 3H), 1.88–1.65 (m, 4H), 1.71 (s, 3H), 1.58 (s, 3H)
14h	prenyl	Me	3-aminopyrrolidine	A	58	232–233	12.31 (br s, 1H), 9.67 (br s, 2H), 7.61 (d, 1H, $J = 8.18$ Hz), 6.51 (d, 1H, $J = 7.14$ Hz), 5.66 (t, 1H, $J = 7.78$ Hz), 4.71–4.47 (m, 2H), 4.16–4.04 (m, 1H), 3.20–3.05 (m, 4H), 2.45 (s, 3H), 2.20–2.03 (m, 2H), 1.76 (s, 3H), 1.61 (s, 3H)
15a	prenyl	Ph	piperazine	A	70	155–156	13.51 (br s, 1H), 9.62 (br s, 1H), 8.01 (d, 1H, $J = 8.08$ Hz), 7.94 (d, 1H, $J = 7.75$ Hz), 7.81–7.56 (m, 5H), 5.90 (t, 1H, $J = 7.68$ Hz), 4.81–4.50 (m, 2H), 3.71–3.55 (m, 4H), 3.11–2.69 (m, 4H), 1.66 (s, 3H), 1.51 (s, 3H)
15b	prenyl	Ph	ethylenediamine	B	59	158–159	12.01 (br s, 1H), 10.17 (br s, 1H), 9.51 (br s, 2H), 7.61–7.35 (m, 6H), 6.14 (d, 1H, $J = 7.27$ Hz), 5.81 (t, 1H, $J = 7.76$ Hz), 4.69–4.37 (m, 2H), 3.15–2.94 (m, 4H), 1.76 (s, 3H), 1.54 (s, 3H)
15c	prenyl	Ph	1-methylpiperazine	A	62	230–231	13.02 (br s, 1H), 7.81–7.53 (m, 6H), 6.37 (d, 1H, $J = 7.68$ Hz), 5.60 (t, 1H, $J = 7.69$ Hz), 4.71–4.48 (m, 2H), 3.32–3.05 (m, 4H), 2.48–2.31 (m, 4H), 2.29 (s, 3H), 1.65 (s, 3H), 1.52(s, 3H)
15d	prenyl	Ph	morpholine	A	57	262–163	12.82 (br s, 1H), 7.74–7.46 (m, 6H), 6.73 (d, 1H, $J = 7.26$ Hz), 5.61 (t, 1H, $J = 7.71$ Hz), 4.67–4.41 (m, 2H), 3.76–3.59 (m, 4H), 3.32–2.98 (m, 4H), 1.62 (s, 3H), 1.48 (s, 3H)
15e	prenyl	Ph	thiomorpholine	B	60	174–175	13.30 (br s, 1H), 7.59–7.28 (m, 6H), 6.66 (d, 1H, $J = 7.25$ Hz), 5.58 (t, 1H, $J = 7.78$ Hz), 4.59–4.28 (m, 2H), 3.64–3.41 (m, 4H), 3.30–2.93 (m, 4H), 1.67 (s, 3H), 1.52 (s, 3H)
15f	prenyl	Ph	3,5-dimethylpiperazine	B	71	230	12.65 (br s, 1H), 10.18 (br s, 1H), 7.74–7.38 (m, 6H), 6.61 (d, 1H, $J = 7.15$ Hz), 5.49 (t, 1H, $J = 7.70$ Hz), 4.81–4.57 (m, 2H), 3.34–2.89 (m, 6H), 1.66 (s, 3H), 1.59 (s, 3H), 1.24 (s, 3H), 1.13 (s, 3H)
15g	prenyl	Ph	pyrrolidine	C	65	235–236	12.78 (br s, 1H), 7.81–7.38 (m, 6H), 6.42 (d, 1H, $J = 6.96$ Hz), 5.69 (t, 1H, $J = 7.72$ Hz), 4.61–4.38 (m, 2H), 3.20–2.79 (m, 4H), 1.84–1.69 (m, 4H), 1.64 (s, 3H), 1.56 (s, 3H)
15h	prenyl	Ph	3-aminopyrrolidine	B	57	157–158	12.86 (br s, 1H), 9.34 (br s, 2H), 7.88–7.61 (m, 6H), 6.71 (d, 1H, $J = 7.75$ Hz), 5.60 (t, 1H, $J = 7.81$ Hz), 4.66–4.39 (m, 2H), 3.96–3,82 (m, 1H), 3.18–3.05 (m, 4H), 2.26–2.04 (m, 2H), 1.63 (s, 3H), 1.50 (s, 3H)

[a] Recrystallization method A: DMSO/ethanol, method B: MeOH/AcOH, method C: MeOH or EtOH/10% HCl.
[b] Yields are those obtained from the replacement step to the final product isolation including hydrolysis.
[c] Decomposition.

lane, and *J*-values were in Hz. Infrared spectra (IR) were obtained on a Jasco FT/IR-300E spectrometer. Mass spectra (MS) were recorded on a Shimadzu-LKB 9000 GC/MS system. High resolution mass spectra (HRMS) were obtained on a JEOL JMS-HX-110A/110A high resolution mass spectrometer. Elemental analyses were performed on a CE instruments model 1110 elemental analyzer. All compounds have analytical results within $\pm 0.4\%$ of their theoretical values. All melting points were uncorrected. When necessary, chemicals were purified according to the reported procedure [15].

4.1. In vitro antibacterial activity

All cultures were stored as frozen stock. For the MICs determinations, a broth microdilution method [16,17] was used to quantitate antibacterial activity for these compounds. The antibacterial activity was determined by agar dilution assay using a multipoint inoculator. The test compounds were dissolved and incorporated by the twofold dilution method in the agar medium. Bacterial inocula, coming from overnight broth and containing 10^7 colony-forming units per point, were inoculated by multipoint inoculator. Bacte-

Table 2
In vitro antibacterial activity of 2-substituted quinolone derivatives

Comp.	Minimum inhibitory concentrations (MICs) μg/ml										
	Gram-positive organisms [a]				Gram-negative organisms [b]						
	Sp		Sf	Sa	Ec	Pa		St	Ko	Ka	Ec P 99
	308 A	77 A				9027	1771 M				
12a	3.2	12.5	6.3	12.5	12.5	25	6.3	12.5	25	12.5	12.5
12b	>25	>25	>25	>25	>25	>25	>25	>25	>25	>25	>25
12c	12.5	6.3	3.2	12.5	12.5	6.3	12.5	25	12.5	25	12.5
12d	6.3	12.5	12.5	6.3	12.5	12.5	6.3	6.3	12.5	12.5	12.5
12e	12.5	12.5	6.3	6.3	25	12.5	6.3	3.2	25	12.5	12.5
12f	6.3	6.3	6.3	12.5	12.5	12.5	25	3.2	25	6.3	12.5
12g	6.3	12.5	12.5	12.5	25	25	6.3	6.1	12.5	6.3	25
12h	12.5	12.5	12.5	12.5	12.5	12.5	6.3	12.5	3.2	12.5	25
13a	6.3	12.5	12.5	6.3	12.5	6.3	6.3	3.1	12.5	3.2	6.3
13b	12.5	6.3	3.2	12.5	6.3	12.5	25	25	25	3.2	12.5
13c	12.5	3.2	12.5	12.5	25	25	6.3	6.3	25	3.2	25
13d	6.3	12.5	25	12.5	12.5	25	25	12.5	25	1.6	25
13e	12.5	12.5	12.5	6.3	25	25	25	12.5	12.5	6.3	25
13f	6.3	6.3	6.3	12.5	25	6.3	12.5	6.3	12.5	3.2	6.3
13g	6.3	6.3	12.5	6.3	25	12.5	25	12.5	3.2	12.5	12.5
13h	6.3	3.2	25	3.2	6.3	12.5	12.5	12.5	12.5	12.5	12.5
14a	3.2	6.3	6.3	0.8	6.3	12.5	6.3	25	25	3.1	12.5
14b	1.6	6.3	6.3	3.2	12.5	6.3	6.3	12.5	6.3	1.6	25
14c	6.3	6.3	3.2	1.6	6.3	12.5	6.3	25	6.3	1.6	25
14d	6.3	6.3	6.3	3.2	25	12.5	25	6.3	12.5	25	12.5
14e	6.3	6.3	6.3	3.2	12.5	25	25	6.3	12.5	12.5	6.3
14f	1.6	3.2	0.8	1.6	25	12.5	6.3	12.5	12.5	12.5	12.5
14g	3.2	6.3	12.5	1.6	6.3	25	25	3.2	6.3	3.2	25
14h	6.3	12.5	6.3	0.8	12.5	12.5	12.5	12.5	6.3	6.3	12.5
15a	6.3	3.2	6.3	1.6	6.3	12.5	6.3	3.2	3.2	6.3	12.5
15b	6.3	6.3	6.3	3.2	12.5	12.5	6.3	25	12.5	3.2	12.5
15c	1.6	3.2	3.2	1.6	25	6.3	6.3	25	25	25	6.3
15d	3.2	3.2	6.3	1.6	25	25	3.2	6.3	12.5	12.5	25
15e	3.2	6.3	6.3	0.8	>25	12.5	12.5	12.5	3.2	12.5	12.5
15f	6.3	6.3	6.3	3.2	25	3.2	25	6.3	3.2	3.2	25
15g	3.2	6.3	6.3	0.8	12.5	25	6.3	12.5	12.5	12.5	6.3
15h	6.3	6.3	3.2	1.6	25	12.5	3.2	12.5	6.3	12.5	12.5
NFLX [c]	1.6	1.6	3.2	0.4	0.01	0.16	0.1	0.02	0.1	0.63	0.4

[a] The screening organisms: Gram-positive bacteria: *Streptococcus pyogenes* 308 A, *Streptococcus pyogenes* 77 A (Sp), *Streptococcus faccium* (Sf), *Staphylococcus aureus* (Sa).

[b] Gram-negative bacteria: *Escherichia coli* 078 (Ec), *Pseudomonas aeruginosa* 9027, *Pseudomonas aeruginosa* 1771 M (Pa), *Salmonella typhimurium* (St), *Klebsiella oxytoca* 1082 E (Ko), *Klebsiella aerogenes* 1522 E (Ka), *Enterobacter cloacae* P 99 (Ec P 99).

[c] The compared drug: Norfloxacin (NPLX).

rial growth was observed after 18 h, of incubation at 37°C. The lowest concentrations of tested compounds that completely inhibited growth were considered to be the minimum inhibitory concentrations (MICs).

4.1.1. 4,5-Difluoro-2-nitrobenzoic acid (**2b**)

A solution of nitric acid (70%, 9.9 g, 110 mmol) in concentrated sulfuric acid (11.4 g, 110 mmol) was added dropwise to a solution of 3,4-difluorobenzoic acid (15.8 g, 100 mmol) in 60 ml of concentrated sulfuric acid at 10°C for 1 h. The reaction mixture was stirred at room temperature (r.t.) for 3 h, and diluted with 120 ml of ice water in an ice-salt bath. The product was collected by filtration, washed with water and cold ethanol. The pale yellow solid was dried at 40°C for 16 h, to give 17.5 g (86%) of **2b**, $R_f = 0.28$ (ethyl acetate, neat); m.p. 158–159°C (Ref. [18], 152–154°C); IR (ν_{max}, KBr): 3310, 1716, 1540, 1438, 1196, 660 cm^{-1}; ^1H NMR (acetone-d_6): δ 10.05 (br s, 1H), 8.14 (dd, 1H, $J = 7.19$ Hz, $J = 3.66$ Hz), 7.93 (dd, 1H, $J = 8.00$ Hz, $J = 4.34$ Hz). ^{13}C NMR (acetone-d_6): δ 164.36, 152.82, 152.03, 146.11, 125.74, 120.25, 115.53.

4.2. General procedure for the preparation of keto esters (**4a–d**)

Thionyl chloride (60.3 mmol) was added to a well stirred suspension of compounds **2a**,**b** (50.2 mmol) and urea (0.3 g) in 40 ml of anhydrous toluene. The reaction mixture was heated in an oil bath at 100°C for 3 h, then cooled to r.t. The mixture of compound **3** (50.2 mmol), magnesium (53.4 mmol), ethanol (165 mmol), carbon tetrachloride (1.1 ml) and 80 ml of anhydrous toluene was stirred at r.t. for 1 h, and refluxed for 1 h. The reaction mixture was cooled to 5°C. The former solution was added to the latter. The resulting reaction mixture was stirred at r.t. for 30 min, and then 20 ml of 10% hydrochloric acid was added. The mixture was extracted with ether, and the combined organic extracts were washed with brine, dried and concentrated at reduced pressure.

4.2.1. Diethyl 2-(2-nitrobenzoyl)malonate (**4a**)

This compound was obtained in 99% yield as a beige liquid (Ref. [19]), $R_f = 0.33$ (hexane/ethyl acetate = 3/1); IR (ν_{max}, $CHCl_3$): 3428, 2964, 1726, 1702, 1455, 1196 cm^{-1}. ^1H NMR ($CDCl_3$): δ 14.53 (br s, 1H), 7.82–7.46 (m, 3H), 7.38 (d, 1H, $J = 7.06$ Hz), 4.18 (q, 2H, $J = 7.12$ Hz), 4.10 (q, 2H, $J = 7.14$ Hz), 1.26 (t, 3H, $J = 7.12$ Hz), 1.11 (t, 3H, $J = 7.14$ Hz). ^{13}C NMR ($CDCl_3$): δ 176.10, 169.76, 166.23, 152.38, 133.92, 130.61, 126.84, 125.13, 119.27, 97.44, 62.46, 61.85, 14.21, 14.06.

4.2.2. Diethyl 2-(4,5-difluoro-2-nitrobenzoyl)malonate (**4b**)

This compound was obtained in 95% yield as a yellow liquid, $R_f = 0.30$ (hexane/ethyl acetate = 3/1); IR (ν_{max}, $CHCl_3$): 3390, 1703, 1687, 1460, 1080, cm^{-1}. ^1H NMR ($CDCl_3$): δ 13.26 (br s, 1H), 8.11 (dd, 1H, $J = 7.09$ Hz, $J = 6.93$ Hz), 7.33 (dd, 1H, $J = 8.69$ Hz, $J = 7.92$ Hz), 4.24 (q, 2H, $J = 7.13$ Hz), 4.16 (q, 2H, $J = 7.12$ Hz), 1.26 (t, 3H, $J = 7.13$ Hz), 1.18 (t, 3H, $J = 7.12$ Hz). ^{13}C NMR ($CDCl_3$): δ 169.41, 168.93, 166.24, 158.10, 152.92, 148.23, 120.32, 118.64, 113.81, 95.73, 62.03, 61.94, 14.02, 13.87; MS (FAB^+): 346 ($M^+ + 1$), 274, 186 (base peak), 154, 137. HRMS: Calc. for 346.0738. Found 346.0734 ($M^+ + 1$).

4.2.3. Ethyl 2-(4,5-difluoro-2-nitrobenzoyl)-3-oxobutanoate (**4c**)

This compound was obtained in 97% yield as a yellow liquid, $R_f = 0.30$ (hexane/ethyl acetate = 5/1); IR (ν_{max}, $CHCl_3$): 3405, 1712, 1639, 1437, 1014 cm^{-1}; ^1H NMR (DMSO-d_6): δ 13.55 (br s, 1H), 8.47 (dd, 1H, $J = 6.99$ Hz, $J = 6.95$ Hz), 7.83 (dd, 1H, $J = 7.77$ Hz, $J = 7.75$ Hz), 3.90 (q, 2H, $J = 7.09$ Hz), 2.45 (s, 3H), 0.86 (t, 3H, $J = 7.09$ Hz). ^{13}C NMR (DMSO-d_6): δ 192.31, 173.66, 165.87, 157.24, 155.03, 149.84, 119.62, 118.56, 113.47, 100.38, 58.92, 24.27, 13.41. MS (FAB^+): 316 ($M^+ + 1$, base peak), 270, 186, 137. HRMS: Calc. for 316.0633. Found 316.0632 ($M^+ + 1$).

4.2.4. Ethyl 2-benzoyl-3-(4,5-difluoro-2-nitrophenyl)-3-oxopropanone (**4d**)

This compound was obtained in 98% yield as a beige liquid, $R_f = 0.33$ (hexane/ethyl acetate = 5/1); IR (ν_{max}, $CHCl_3$): 3378, 1711, 1604, 1541, 1411, 1148 cm^{-1}. ^1H NMR (DMSO-d_6): δ 13.60 (br s, 1H), 8.62 (dd, 1H, $J = 7.16$ Hz, $J = 7.13$ Hz), 8.15–8.05 (m, 3H), 7.95 (dd, 1H, $J = 7.93$ Hz, $J = 7.92$ Hz), 7.67–7.60 (m, 2H), 3.94 (q, 2H, $J = 7.15$ Hz), 0.89 (t, 3H, $J = 7.15$ Hz); ^{13}C NMR (DMSO-d_6): δ 193.61, 173.19, 170.75, 158.04, 152.55, 150.26, 136.43, 133.83, 133.36, 129.87, 129.12, 127.63, 120.24, 118.75, 112.96, 95.82, 60.08, 13.97. MS (FAB^+): 378 ($M^+ + 1$, base peak), 332, 274, 193, 105, 77. HRMS: Calc. for 378.0789. Found 378.0789 ($M^+ + 1$).

4.2.5. 4-Hydroxy-1,2-dihydro-2-oxo-3-quinolinecarboxylicacid ethyl ester (**5a**)

To a solution of keto diester **4a** (3.1 g, 10 mmol) in dioxane (45 ml) was added 20% aqueous sodium hydroxide (10 ml) and 10% palladium-on-charcoal (0.4 g) at r.t. The mixture was stirred for 20 min, and then added dropwise to a solution of sodium borohydride (0.7 g, 18.5 mmol) in water (5 ml). The reaction mixture was stirred at r.t. for 30 min, and filtered through Celite. The filtrate was concentrated to remove dioxane

and then the residue was acidified with 10% hydrochloric acid to give pale yellow solid, which was recrystallized from ethanol to give 1.6 g (70%) of **5a**, $R_f = 0.23$ (chloroform/methanol = 10/1); m.p. 300°C (dec, Ref. [20]; 304°C); IR (ν_{max}, KBr): 3320–3084, 1711, 1412, 1144 cm^{-1}. ^1H NMR (DMSO-d_6): δ 12.64 (br s, 1H), 8.22–8.01 (m, 3H), 7.95 (d, 1H, $J = 6.97$ Hz), 4.26 (q, 2H, $J = 7.11$ Hz), 1.30 (t, 3H, $J = 7.11$ Hz). ^{13}C NMR (DMSO-d_6): δ 172.31, 171.43, 164.24, 151.19, 129.20, 126.36, 125.74, 120.21, 105.13, 99.04, 59.85, 13.94.

According to the same procedure, 6,7-difluoro-4-hydroxy-1,2-dihydro-2-oxo-3-quinoline carboxylic acid ethyl ester (**5b**) was prepared. Compound **5b** was obtained in 72% yield from **4b**, $R_f = 0.26$ (chloroform/methanol = 7/1); m.p. 280°C (dec); IR (ν_{max}, KBr): 3270–3030, 1649, 1579, 1188 cm^{-1}. ^1H NMR (DMSO-d_6): δ 12.86 (br s, 1H), 8.10 (dd, 1H, $J = 9.34$ Hz, $J = 9.17$ Hz), 7.71 (dd, 1H, $J = 7.01$ Hz, $J = 6.79$ Hz), 4.44 (q, 2H, $J = 7.13$ Hz), 1.40 (t, 3H, $J = 7.13$ Hz). ^{13}C NMR (DMSO-d_6): δ 168.24, 165.68, 162.87, 152.05, 148.55, 143.92, 117.24, 112.91, 109.89, 101.94, 61.86, 14.36; MS (m/e): 269 (M^+), 239 (base peak), 171, 143, 115; *Anal*. Calc. for $C_{12}H_9N$: C, 53.54; H, 3.37; N, 5.20. Found C, 55.36; H, 3.51; N, 5.39%.

*4.2.6. 6,7-Difluoro-1-hydroxy-2-methyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid ethyl ester (**6a**)*

A solution of the diketo ester **4c** (3.2 g, 10.2 mmol) in 80 ml of ethanol was hydrogenated over 10% palladium-on-charcoal (0.6 g) under atmospheric pressure at r.t. for 3 h. The reaction mixture was filtered through Celite and evaporated to give a pale yellow solid, which was recrystallized from ethanol to give 2.2 g (78%) of **6a**, $R_f = 0.33$ (chloroform/methanol = 10/1); m.p. 202°C; IR (ν_{max}, KBr): 3450, 1717, 1605, 1475, 1110 cm^{-1}. ^1H NMR (DMSO-d_6): δ 12.33 (br s, 1H), 8.04 (dd, 1H, $J = 10.07$ Hz, $J = 8.44$ Hz), 7.91 (dd, 1H, $J = 7.09$ Hz, $J = 6.77$ Hz), 4.26 (q, 2H, $J = 7.10$ Hz), 2.42 (s, 3H), 1.30 (t, 3H, $J = 7.10$ Hz). ^{13}C NMR (DMSO-d_6): δ 168.04, 164.87, 152.52, 149.10, 147.37, 143.68, 135.24, 119.88, 110.94, 102.36, 58.95, 13.67, 12.21; MS (m/e): 283 (M^+). HRMS: Calc. for 283.0707. Found 283.0727 (M^+).

According to the same procedure, 6,7-difluoro-1-hydroxy-2-phenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid ethyl ester (**6b**) was prepared. Compound **6b** was obtained in 71% yield from **4d**, $R_f = 0.26$ (chloroform/methanol = 10/1); m.p. 194–195°C; IR (ν_{max}, KBr): 3414, 1720, 1608, 1477, 1102 cm^{-1}. ^1H NMR (DMSO-d_6): δ 12.26 (br s, 1H), 8.17 (dd, 1H, $J = 8.61$ Hz, $J = 8.59$ Hz), 7.89 (dd, 1H, $J = 7.72$ Hz, $J = 7.72$ Hz), 7.63–7.53 (m, 5H), 3.96 (q, 2H, $J = 7.11$ Hz), 0.90 (t, 3H, $J = 7.11$ Hz). ^{13}C NMR (DMSO-d_6): δ 169.85, 164.42, 154.35, 150.87, 150.45, 148.69, 145.31, 136.82, 130.14, 129.46, 128.57, 127.65, 121.73, 115.18, 112.58, 104.42, 59.95, 13.07; MS (m/e): 345 (M^+). HRMS: Calc. for 329.0863. Found 329.0847 (M^+).

*4.2.7. 6,7-Difluoro-2-methyl-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid ethyl ester (**7a**)*

To a solution of prenyl bromide (1.9 g, 12.7 mmol) and compound **6a** (3.0 g, 10.6 mmol) in *N*,*N*-dimethylformamide (36 ml) potassium carbonate (7.3 g, 53.0 mmol) was added. The reaction mixture was heated at 80–90°C for 6 h. The reaction mixture was poured into ice-water (120 g) and stirred for 30 min. The solution was extracted with chloroform (3 × 100 ml), washed with H_2O and brine. The organic layer was dried ($MgSO_4$), filtered and concentrated under vacuum. The resulting solid was filtered and washed with water and then recrystallized from ethanol to give 2.5 g, (71%) of **7a** as pale yellow crystal. $R_f = 0.34$ (chloroform/methanol = 10/1); m.p. 99–100°C; IR (ν_{max}, KBr): 1726, 1638, 1464, 1155 cm^{-1}. ^1H NMR (DMSO-d_6): δ 8.01 (dd, 1H, $J = 8.60$ Hz, $J = 8.60$ Hz), 7.74 (dd, 1H, $J = 7.75$ Hz, $J = 7.75$ Hz), 5.62 (t, 1H, $J = 7.75$ Hz), 4.82–4.69 (m, 2H), 4.26 (q, 2H, $J = 7.12$ Hz), 2.51 (s, 3H), 1.79 (s, 3H), 1.69 (s, 3H), 1.27 (t, 3H, $J = 7.12$ Hz). ^{13}C NMR (DMSO-d_6): δ 172.27, 160.26, 152.05, 148.41, 141.68, 137.41, 123.58, 119.04, 116.23, 115.58, 114.93, 103.51, 61.16, 50.59, 23.57, 21.60, 17.43, 13.96.

According to the same procedure, 6,7-difluoro-2-phenyl-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid ethyl ester (**7b**) was prepared. Compound 7**b** was obtained in 72% yield from **6b** as beige solid. $R_f = 0.38$ (chloroform/methanol = 10/1); m.p. 89–90°C; IR (ν_{max}, KBr): 1723, 1648, 1485, 1239 cm^{-1}. ^1H NMR (DMSO-d_6): δ 7.92 (dd, 1H, $J = 8.43$ Hz, $J = 8.43$ Hz), 7.49 (dd, 1H, $J = 7.61$ Hz, $J = 7.61$ Hz), 7.38–7.18 (m, 5H), 5.60 (t, 1H, $J = 7.56$ Hz), 4.71–4.60 (m, 2H), 4.29 (q, 2H, $J = 7.12$ Hz), 1.81 (s, 3H), 1.70 (s, 3H), 1.27 (t, 3H, $J = 7.12$ Hz). ^{13}C NMR (DMSO-d_6): δ 172.92, 163.28, 153.54, 148.49, 141.98, 140.07, 138.48, 137.41, 130.27, 127.67, 125.58, 124.03, 123.87, 118.39, 117.62, 114.43, 104.08, 102.01, 61.19, 53.31, 23.64, 21.60, 13.94.

*4.2.8. 6,7-Difluoro-2-methyl-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid (**8a**)*

A suspension of **7a** (1.2 g, 3.6 mmol) in 50 ml of 2*N* aqueous sodium hydroxide was refluxed for 3 h. The reaction mixture was cooled and acidified with 10% hydrochloric acid. The white solid was collected by filtration, washed with water and dried to give 1.0 g (94%) of **8a**. $R_f = 0.20$ (chloroform/methanol = 3/1); m.p. 300°C (dec); IR (ν_{max}, KBr): 3468, 1642, 1487, 1230 cm^{-1}. ^1H NMR (DMSO-d_6): δ 12.23 (br s, 1H), 8.16 (dd, 1H, $J = 7.87$ Hz, $J = 7.84$ Hz), 8.10 (dd, 1H, $J = 7.61$ Hz, $J = 7.58$ Hz), 5.66 (t, 1H, $J = 7.42$ Hz), 4.71–4.31 (m, 2H), 2.50 (s, 3H), 1.74 (s, 3H), 1.66 (s,

3H). ^{13}C NMR (DMSO-d_6): δ 173.14, 167.44, 158.20, 149.61, 146.21, 144.89, 138.42, 127.13, 118.62, 113.75, 110.27, 103.45, 50.41, 22.94, 21.03, 16.71.

4.2.9. 6,7-Difluoro-2-phenyl-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid (**8b**)

To a solution of **7b** (0.6 g, 1.5 mmol) in tetrahydrofuran/water (v/v, 3:1, 30 ml) was added lithium hydroxide (0.2 g, 8.4 mmol) in tetrahydrofuran (8 ml). The mixture was stirred at r.t. for 18 h. Reaction mixture was acidified with 10% hydrochloric acid. The white solid was collected by filtration, washed with water and dried to give (0.5 g, 92%) of **8b**. $R_f = 0.31$ (chloroform/methanol = 10/1); m.p. 298°C (dec); IR (ν_{max}, KBr): 3348, 1651, 1544, 1126 cm^{-1}. ^{1}H NMR (DMSO-d_6): δ 13.02 (br s, 1H), 8.12 (dd, 1H, $J = 7.91$ Hz, $J = 7.86$ Hz), 8.96 (dd, 1H, $J = 7.55$ Hz, $J = 7.49$ Hz), 7.51–7.40 (m, 5H), 5.58 (t, 1H, $J = 7.64$ Hz), 4.87–4.72 (m, 2H), 1.81 (s, 3H), 1.73 (s, 3H). ^{13}C NMR (DMSO-d_6): δ 173.35, 168.63, 160.24, 153.74, 152.42, 148.56, 142.37, 139.05, 137.41, 129.43, 128.60, 126.11, 124.61, 123.01, 118.27, 117.71, 114.62, 104.84, 52.13, 23.57, 21.61.

4.2.10. Diacetoxy[[[6,7-difluoro-2-methyl-1,4-dihydro-4-oxo-quinolin-3-yl]carbonyl]oxy]borane (**9a**)

To solution of triacetoxyborate in acetic anhydride, which was prepared by stirring the mixture of boric acid (0.74 g, 12 mmol) and acetic anhydride (7.8 g, 76 mmol) at 80°C for 1 h, was added compound **6a** (2.0 g, 7.0 mmol), and the mixture was stirred at 100°C for 3 h, and then left standing at r.t. The precipitates were collected and washed with isopropyl ether to afford **9a** (2.0 g, 78%) as pale yellow crystals. This compound was recrystallized from ethanol. $R_f = 0.41$ (ethyl acetate, neat); m.p. 195°C; IR (ν_{max}, KBr): 3208, 1763, 1715, 1593, 1222 cm^{-1}. ^{1}H NMR (DMSO-d_6): δ 12.60 (br s, 1H), 8.29 (dd, 1H, $J = 7.95$ Hz, $J = 7.95$ Hz), 8.14 (dd, 1H, $J = 7.62$ Hz, $J = 7.62$ Hz), 2.24 (s, 3H), 1.90 (s, 3H), 1.88 (s, 3H). ^{13}C NMR (DMSO-d_6): δ 178.64, 172.86, 167.28, 159.49, 158.21, 150.92, 142.61, 120.42, 113.16, 112.72, 104.26, 25.11, 24.91, 17.04, 16.81.

4.2.11. 6,7-Difluoro-2-methyl-1,4-dihydro-4-oxo-quinolinecarboxylic acid (**10a**)

A suspension of **6a** (1.4 g, 4.9 mmol) in 50 ml of 2 N aqueous sodium hydroxide was refluxed for 3 h. The reaction mixture was cooled and acidified with 10% hydrochloric acid. The white solid was collected by filtration, washed with water and dried to give 1.1 g (94%) of **10a**. $R_f = 0.22$ (chloroform/methanol = 3/1); m.p. 310°C (dec); IR (ν_{max}, KBr): 3447, 1636, 1465, 1227, 697 cm^{-1}. ^{1}H NMR (DMSO-d_6): δ 12.48 (br s, 1H), 9.65 (br s, 1H), 8.25 (dd, 1H, $J = 7.76$ Hz, $J = 7.76$ Hz), 8.03 (dd, 1H, $J = 7.31$ Hz, $J = 7.30$ Hz), 2.45 (s, 3H). ^{13}C NMR (DMSO-d_6): δ 173.12, 167.43, 158.27, 149.63, 146.16, 144.82, 127.18, 115.64, 113.76, 110.22, 16.73; MS (m/e): 239 (M^+); HRMS: Calc. for 239.0394, Found 239.0403 (M^+). *Anal.* Calc. for $C_{11}H_7N_1$: C, 55.24; H, 2.95; N, 5.86. Found C, 56.63; H, 3.02; N, 5.68%.

According to the same procedure, 6,7-difluoro-2-phenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid (**10b**) was prepared. Compound **10b** was obtained in 92% yield from **6b**. $R_f = 0.25$ (chloroform/methanol = 3/1); m.p. 310°C (dec); IR (ν_{max}, KBr): 3447, 1636, 1465, 1227, 697 cm^{-1}. ^{1}H NMR (DMSO-d_6): δ 13.02 (br s, 1H), 9.84 (br s, 1H), 8.12 (dd, 1H, $J = 7.91$ Hz, $J = 7.86$ Hz), 8.96 (dd, 1H, $J = 7.55$ Hz, $J = 7.49$ Hz), 7.51–7.40 (m, 5H). ^{13}C NMR (DMSO-d_6): δ 172.85, 167.37, 166.12, 158.41, 149.83, 147.21, 131.66, 130.82, 129.31, 128.53, 127.68, 126.91, 124.20, 116.75, 112.81, 105.93. *Anal.* Calc. for $C_{16}H_9N_1$: C, 63.79; H, 3.01; N, 4.65. Found C, 63.99; H, 3.12; N, 4.51%.

4.2.12. Difluoro[[[6,7-difluoro-2-methyl-1,4-dihydro-4-oxo-quinolin-3-yl]carbonyl]oxo]borane (**11a**)

A mixture of 6,7-difluoro-2-methyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acid **10a** (1.2 g, 5.0 mmol) and 54 wt.% tetrafluoroboric acid (8 ml) in water was heated at 100°C for 3 h, and then left standing at r.t. The precipitates were collected by filtration and washed with isopropyl ether to afford **11a** (1.1 g, 78%) as beige crystals. This compound was recrystallized from ethanol. $R_f = 0.46$ (ethyl acetate, neat); m.p. 195–198°C; IR (ν_{max}, KBr): 3225, 1755, 1705, 1421, 1062 cm^{-1}. ^{1}H NMR (DMSO-d_6): δ 12.48 (br s, 1H), 8.37 (dd, 1H, $J = 7.71$ Hz, $J = 7.71$ Hz), 8.08 (dd, 1H, $J = 7.26$ Hz, $J = 7.26$ Hz), 2.28 (s, 3H). ^{13}C NMR (DMSO-d_6): δ 174.26, 161.29, 159.48, 157.27, 151.56, 147.74, 142.61, 121.52, 112.92, 105.83, 16.72.

According to the same procedure, difluoro[[[6,7-difluoro-2-phenyl-1,4-dihydro-4-oxo-quinolin-3-yl]carbonyl] oxy]borane (**11b**) was prepared. Compound **11b** was obtained in 71% yield from **10b**. $R_f = 0.26$ (ethyl acetate, neat); m.p. 162–163°C; IR (ν_{max}, KBr): 3241, 1739, 1685, 1531, 1148 cm^{-1}. ^{1}H NMR (DMSO-d_6): δ 12.34 (br s, 1H), 8.29 (dd, 1H, $J = 8.62$ Hz, $J = 8,62$ Hz), 7.91 (dd, 1H, $J = 7.63$ Hz, $J = 7.63$ Hz), 7.51–7.28 (m, 5H). ^{13}C NMR (DMSO-d_6): δ 172.08, 165.49, 159.28, 149.33, 148.27, 147.12, 142.86, 131.22, 129.46, 128.15, 124.16, 122.01, 114.64, 112.56, 105.48, 104.26.

4.3. General procedure for the preparation of 7-substituted 6-fluoro-2-(methyl or phenyl)-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids (**12a–h** and **13a–h**)

A mixture of **9a** or **11a**,**b** (3.3 mmol) and appropriate amine (10.0 mmol) in 20 ml of acetonitrile was stirred at r.t. for 6–10 h, under nitrogen atmosphere. The reaction mixture was evaporated under reduced pres-

sure. The residue was treated with water, and then the resulting precipitate was collected by filtration, washed with water and recrystallized from dimethyl sulfoxide/ethanol, methanol/acetic acid, methanol or ethanol/10% hydrochloric acid to afford **12a**–**h** and **13a**–**h**, respectively.

*4.4. General procedure for the preparation of 7-substituted 6-fluoro-2-(methyl or phenyl)-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids (**14a**–**h** and **15a**–**h**)*

A mixture of **8a**,**b** (2.5 mmol) and appropriate amine (10.0 mmol) in 20 ml of pyridine was heated at 100°C for 6–36 h, under nitrogen atmosphere. The reaction mixture was evaporated under reduced pressure. The residue was treated with water, and then the resulting precipitated was collected by filtration, washed with water and recrystallized from dimethyl sulfoxide/ethanol, methanol/acetic acid, methanol or ethanol /10% hydrochloric acid to afford **14a**–**h** and **15a**–**h**, respectively. Their yields, m.p., recrystallization method and ^{1}H NMR (DMSO-d_6) data are given in Table 1.

5. Conclusion

In conclusion, a simple and versatile synthesis of *N*-prenyl 2-substituted quinolone derivatives, i.e. 7-substituted 6-fluoro-2-(methyl or phenyl)-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids (**2a**–**h**, **3a**–**h**) and 7-substituted 6-fluoro-2-(methyl or phenyl)-1-prenyl-1,4-dihydro-4-oxo-3-quinolinecarboxylic acids (**4a**–**h**, **5a**–**h**) has been described. This synthesis is also suitable for large scale preparation. The synthetic strategies involve the use of well known keto ester condensation of benzoyl chloride and reductive cyclization of intermediates (**4a**–**d**) to afford 4-hydroxyquinolones (**5a**,**b**) or 2-substituted quinolone (**6a**,**b**) derivatives. We have found that the *N*-prenyl 2-substituted (methyl or phenyl) quinolone derivatives exhibit the improved antibacterial activity for the Gram-positive organisms in vitro, presumably due to the increased lipophilicity and enhanced self-association. In particular, the 6-fluoro-2-methyl-1-prenyl-1,4-dihydro-7-(3,5-dimethylpiperazinyl)-4-oxo-3-quinolinecarboxylic acid (**14f**) exhibited the most potent antibacterial activity against Gram-positive bacteria among the total 32 derivatives.

Acknowledgements

This work has been supported by the Basic Science Research Institute Program (BSRI-99-3433), Ministry of Education, Republic of Korea. The structure analyses of the compounds were partially supported by Korea Basic Science Institute.

References

[1] G.Y. Lesher, E.D. Forelich, M.D. Gruet, J.H. Bailey, R.P. Brundage, 1,8-Naphthyridine derivatives. A new class of chemotherapeutic agents, J. Med. Pharm. Chem. 5 (1962) 1063–1065.
[2] K. Drlica, X. Zhao, DNA gyrase, topoisomerase IV, and the 4-quinolones, Microbiol. Mol. Biol. Rev. 61 (1997) 377–392.
[3] J.F. Barrett, J.A. Sutcliffe, T.D. Gootz, In vitro assays used to measure the activity of topoisomerases, Antimicrob. Agents Chemother. 34 (1990) 1–7.
[4] P.B. Fernandes, Quinolones, Proceedings of an International Telesymposium, J.R. Prous Science Publishers, Barcelona, Spain, 1989.
[5] D.R. White, L.C. Davenport, Chapter 12. Antibacterial agents, Annu. Rep. Med. Chem. 25 (1989) 109–118.
[6] J. Segawa, M. Kitano, K. Kazuno, M. Matsuoka, I. Sh- irahase, M. Ozaki, M. Matsuda, Y. Tomii, M. Kise, Studies on pyridonecarboxylic acids. 1. Synthesis and antibacterial evaluation of 7-substituted-6-halo-4-oxo-4H-[1,3]thiazeto[3,2-*a*]quino-line-3-carboxylic acids, J. Med. Chem. 35 (1992) 4727–4738.
[7] M. Cruciani, D. Bassetti, The fluoroquinolones as treatment for infections caused by Gram-positive bacteria, J. Antimicrob. Chemother. 33 (1994) 402–417.
[8] H.M. Blumberg, D. Rimland, D.J. Carroll, P. Terry, I.K. Wachsmuth, Rapid development of ciprofloxacin resistance in methicillin-susceptible and -resistant *Staphylococcus aureus*, J. Infect. Dis. 163 (1991) 1279–1285.
[9] L.J.V. Piddock, New quinolones and Gram-positive bacteria, Antimicrob. Agents Chemother. 38 (1994) 163–169.
[10] J. Frigola, D. Vano, A. Torrens, A. Gomez-Gomar, E. Ortega, S. Garcia-Granda, 7-Azetidinylquinolones as antibacterial agents. 3. Synthesis, properties and structure–activity relationships of the stereoisomers containing a 7-(3-amino-2-methyl-1-azetidinyl), Moiety J. Med. Chem. 38 (1995) 1203–1215.
[11] T. Okada, T. Tsuji, T. Tsushima, T. Yoshida, S. Matsuura, Synthesis and antibacterial activities of novel dihydrooxazine and dihydrothiazine ring-fused tricyclic quinolonecarboxylic acids: 9-fluoro-3-methylene-10-(4-methylpiperazin-1-yl)-7-oxo-2,3-dihy-dro-7H-pyrido[1,2,3-*de*][1,4]benzoxazine-6-carboxylic acids and its 1-thia congener, J. Heterocyclic Chem. 28 (1991) 1061–1065.
[12] Y. Inoue, H. Kondo, M. Taguchi, Y. Jinbo, F. Sakamoto, G. Tsukamoto, Synthesis and antibacterial activity of thiazolopyrazine-incorporated tetracyclic quinolone antibacterials, J. Med. Chem. 37 (1994) 586–592.
[13] Y. Jinbo, H. Kondo, M. Taguchi, Y. Inoue, F. Sakamoto, G. Tsukamoto, Synthesis and antibacterial activity of thiazolopyrazine-incorporated tetracyclic quinolone antibacterial agents, J. Med. Chem. 37 (1994) 2791–2796.
[14] D.T.W. Chu, P.B. Fernandes, Recent developments in the field of quinolone antibacterial agents, in: B. Testa (Ed.), Advances in Drug Research, vol. 21, Academic Press, New York, 1991, p. 39.
[15] D.D. Perrin, L.F. Armarego, D.R. Perrin, Purification of Laboratory Chemicals, 2nd edn., Pergamon Press, New York, 1980.
[16] S. Goto, K. Jo, T. Kawakita, N. Kosakai, S. Mitsuhashi, T. Nishino, N. Ohsawa, H. Tanami, About the re-revised method of determination of MIC, Chemotherapy 29 (1981) 76–79.

[17] National Committee for Clinical Laboratory Standards, Methods for dilution antimicrobial susceptibility tests for bacteria grown aerobically; approved standard, National Committee for Clinical Laboratory Standards, Villanova, PA, 1985.

[18] B. Fertel, Lawrence, C. Lin Henry, Process for the preparation of 4,5-difluoroanthranilic acid from 4,5-difluorophthalic anhydride or 4,5-difluorophthalic acid. US Patent 4 994 606, US Appl. (1991) 439,228; Chem. Abstr. 115 (1991) 71130w.

[19] D. Sicker, The regio- and diastereoselective tautomerization of (-(2-nitrobenzoyl)-1,3-dicarbonyl compounds, Pharmazie 46 (1991) 604 Chem. Abstr. 116 (1992) 12–7949k.

[20] R. Johannes, S.A. Reza, B. Andreas, M. Michael, Acetylenchemie 7. Mitt. [1]: mechanistische studien zur synthese von pyrano[3,2-c]chinolin-alkaloiden aus 4-hydroxychinolin-2onen und 3-chlor-3methylbut-1-in, Monatsh. Chem. 119 (1988) 781–791.